Remarks

The courtesies extended to Drs. Sharpless and Fokin, and their representative during the interview held on 23 January 2008 is gratefully acknowledged.

The present invention is based on the discovery that Cu(I) is a surprisingly effective catalyst for the cycloaddition under mild conditions of an organic azide and a terminal alkyne to form a 1,4-disubstitut4ed 1,2,3-triazole. In the presence of catalytic amounts of Cu(I) this cycloaddition reaction is effective at ambient temperature.

During the aforementioned interview Drs. Sharpless and Fokin gave an overview of the involved reaction as well as sources of Cu(I) catalyst, discussed many diverse applications for the invention, and illustrated a wide variety of 1,4-substituents possible for the formed 1,2,3-triazoles. It was also pointed out that substituents in the organic azide and the terminal alkyne are not critical and do not interfere with the cycloaddition reaction. A copy of the computer presentation made to the Examiner, reformatted for better legibility as hard copy, is attached as Appendix A to this document.

In order to obviate the outstanding rejections of claims 1-30 based on 35 U.S.C. 112, first and second paragraphs, these claims are canceled without prejudice to expedite the further prosecution of this application. In lieu thereof, new claims 31-36, inclusive, are presented, and are presently under consideration. Because of the cancellation of claims 1-30, inclusive, no additional claim fee is deemed to be due inasmuch as claims 31-36 include only one independent claim and five claims dependent thereon directly or indirectly. Claims 31-36 are believed to satisfy all requirements of 35 U.S.C. 112, first and second paragraphs.

Claims 31-36 are narrower in scope, do not introduce new matter, and are fully supported by the specification.

In particular, support for claim 31 can be found, *inter alia,* in the specification at page 3, lines 22-27; at page 4, lines 4-8 and 30-31; at page 11, lines 6-12; and at page 16, lines 1-5.

Support for claim 32 is found in the specification, *inter alia*, at page 9, lines 1-2 and lines 21-22; and at page 12, lines 1-9.

Support for claim 33 is found in the specification, *inter alia*, at page 4, lines 31-32; and at page 5, lines 1-6.

Support for claim 34 is found in the specification, *inter alia*, at page 3, lines 28-30; and at page 8, lines 24-26.

Support for claim 35 is found in the specification, *inter alia*, at page 8, line 27.

Support for claim 36 is found in the specification at page 11, lines 23-25.

Previous claims 1-30 have not been rejected based on prior art. The references cited to show the state of the art have been studied with interest but do not vitiate the patentability of the claimed invention. The present application also has a priority date of 30 May 2002 which antedates U.S. Publication No. 2007/0224695 A1. Newly presented claims 31-36, inclusive, are narrower in scope than those originally presented, thus a new search is not necessary. Inasmuch as claims 31-36 are believed to place this application in condition for allowance, entry of this amendment, and early passing of this application to issue is solicited.

Respectfully submitted,

January 24, 2008

By _____
Talivaldis Cepuritis (Reg. No. 20,818)

OLSON & CEPURITIS, LTD.
20 North Wacker Drive
36th Floor
Chicago, Illinois 60606
(312) 580-1180

Attachment: Appendix A (32 pages)

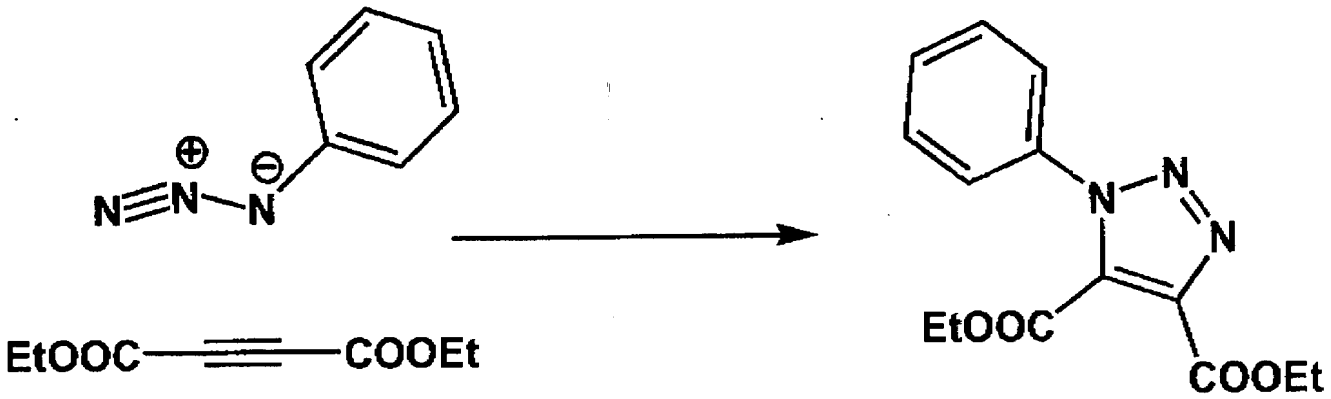
CERTIFICATE OF FACSIMILE TRANSMISSION

I hereby certify that this AMENDMENT AND RESPONSE UNDER RULE 116 is being transmitted by facsimile transmission to Fax No. 571-273-8300 on January 24, 2008.

_____
Talivaldis Cepuritis (Reg. No. 20,818)

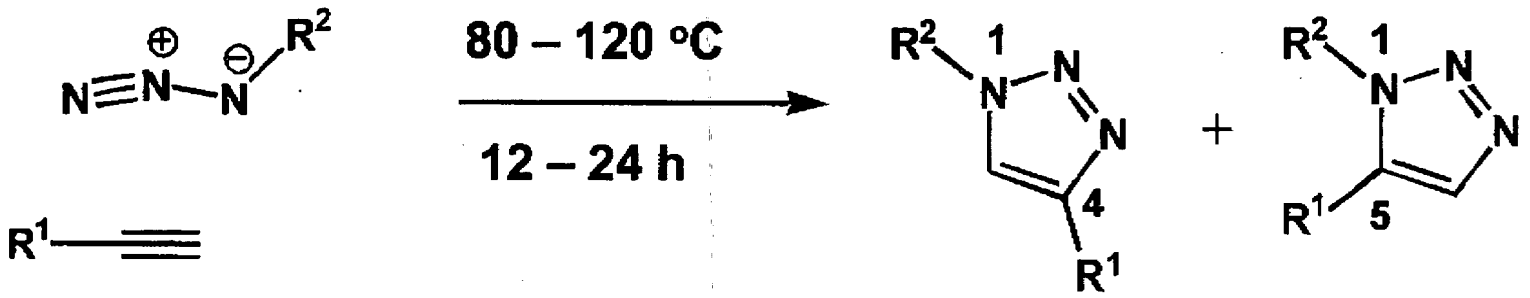
APPENDIX A

Alkynes and Organic Azides: Synthesis of 1,2,3-Triazoles



A. Michael, *J. Prakt Chem.* 1893, *48*, 94.

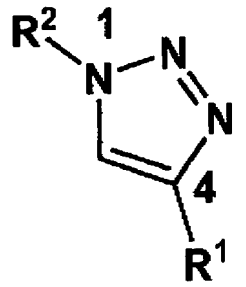
Azide-Alkyne Cycloaddition: What is Missing?

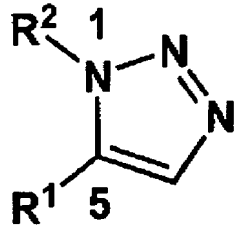


ca. 1 : 1

However, the desired triazole-forming cycloaddition requires elevated temperatures or highly activated dipolarophiles, and usually results in a mixture of the 1,4- and 1,5-regioisomers.

1,2,3-Triazoles





pK_{NH+} = 1.25 (R^1 = H; R^2 = CH_3)

- Very stable to oxidation, reduction, and hydrolysis
- High dipoles (ca. 4.7 – 5.2 Debye)
- Weakly basic (N-2 and N-3 H-bond acceptors)
- Favorable metabolism and toxicology profiles
- Good ligands for metals
- Yet barely utilized

Organic Azides: a Unique Reactivity Manifold

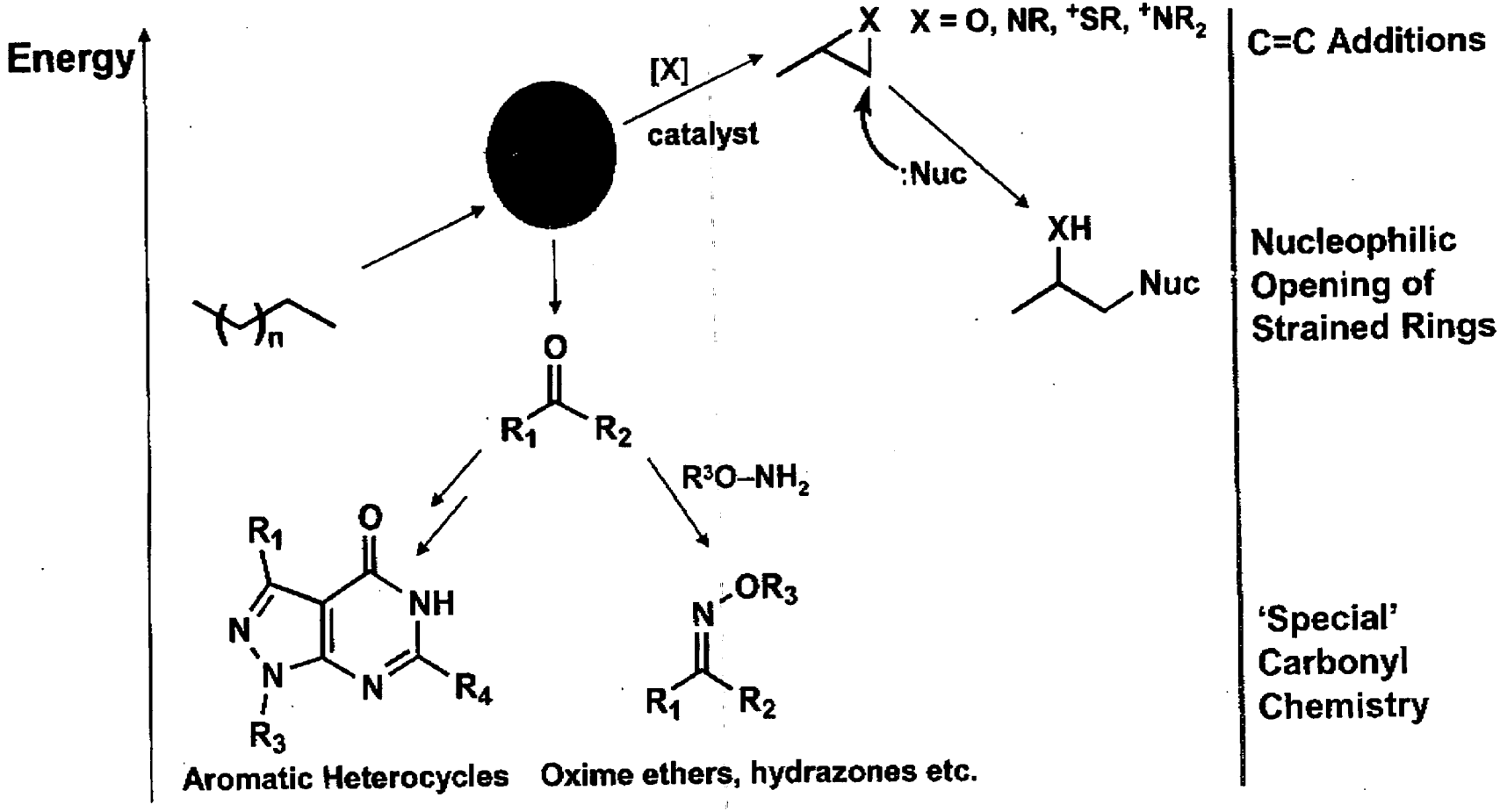
- Very **energetic**
- Yet most are **stable** and can be handled safely if simple precautions are observed
- Virtually **inert** to most other functionalities (certainly bio-orthogonal)

Saxon E., Bertozzi C.R.: Cell Surface Engineering by a Modified Staudinger Reaction. *Science* 2000, 287:2007-2010.

Kolb H.C., Finn M.G., Sharpless K.B.: Click chemistry: Diverse chemical function from a few good reactions. *Angew. Chem. Int. Ed.* 2001, 40:2004-2021.

Click Chemistry:

Modular synthesis using spring-loaded reactions

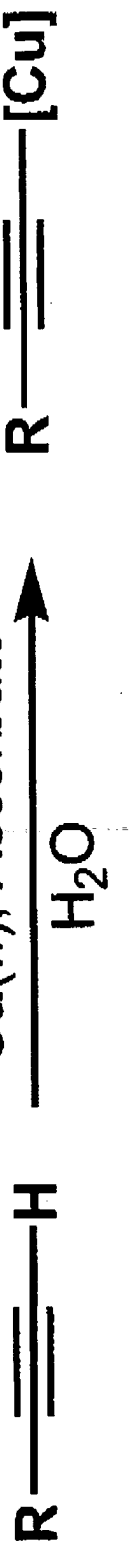


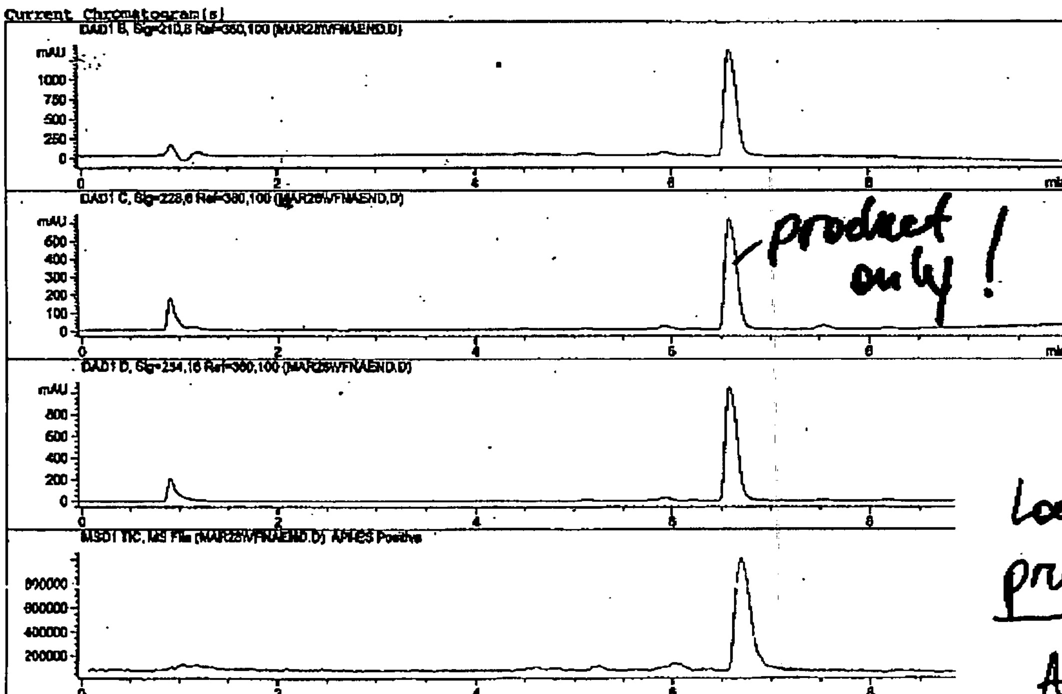
Kolb H.C.; Finn M.G.; Sharpless K.B. Angew. Chem. Int. Ed. **2001**, *40*, 2004-2021

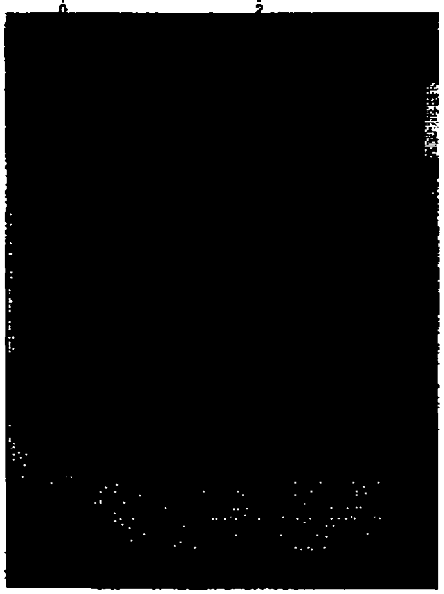
Organic Azides: a Unique Reactivity Manifold

- Very **energetic**
- Yet most are **stable** and can be handled safely if simple precautions are observed
- Virtually **inert** to most other functionalities (certainly bio-orthogonal)

In situ generation of Reactive Copper(I) Acetylides

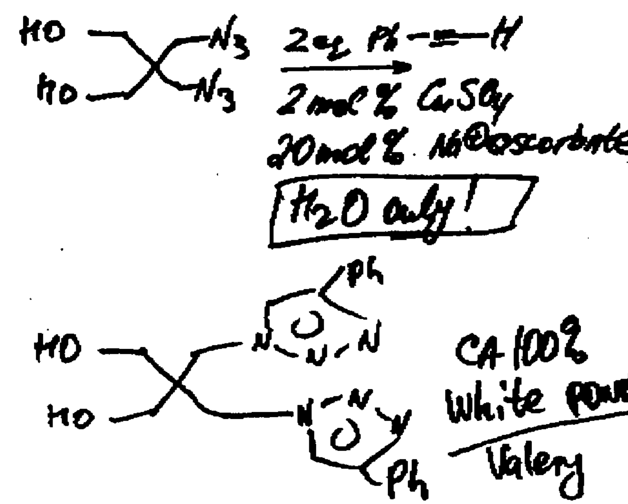






Looks like we have the process!

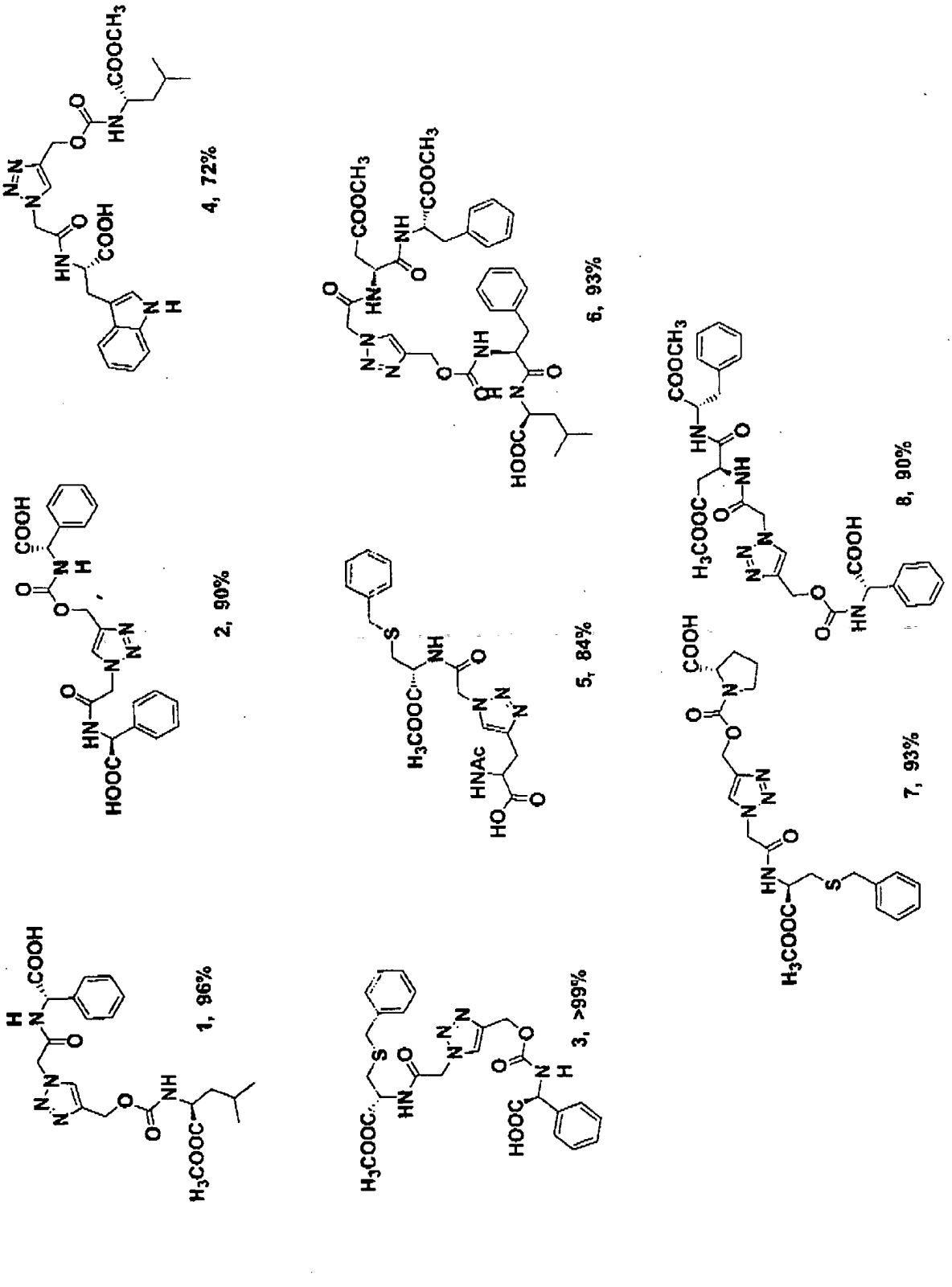
Attached is the LCMS of the rxn after 8 hrs.



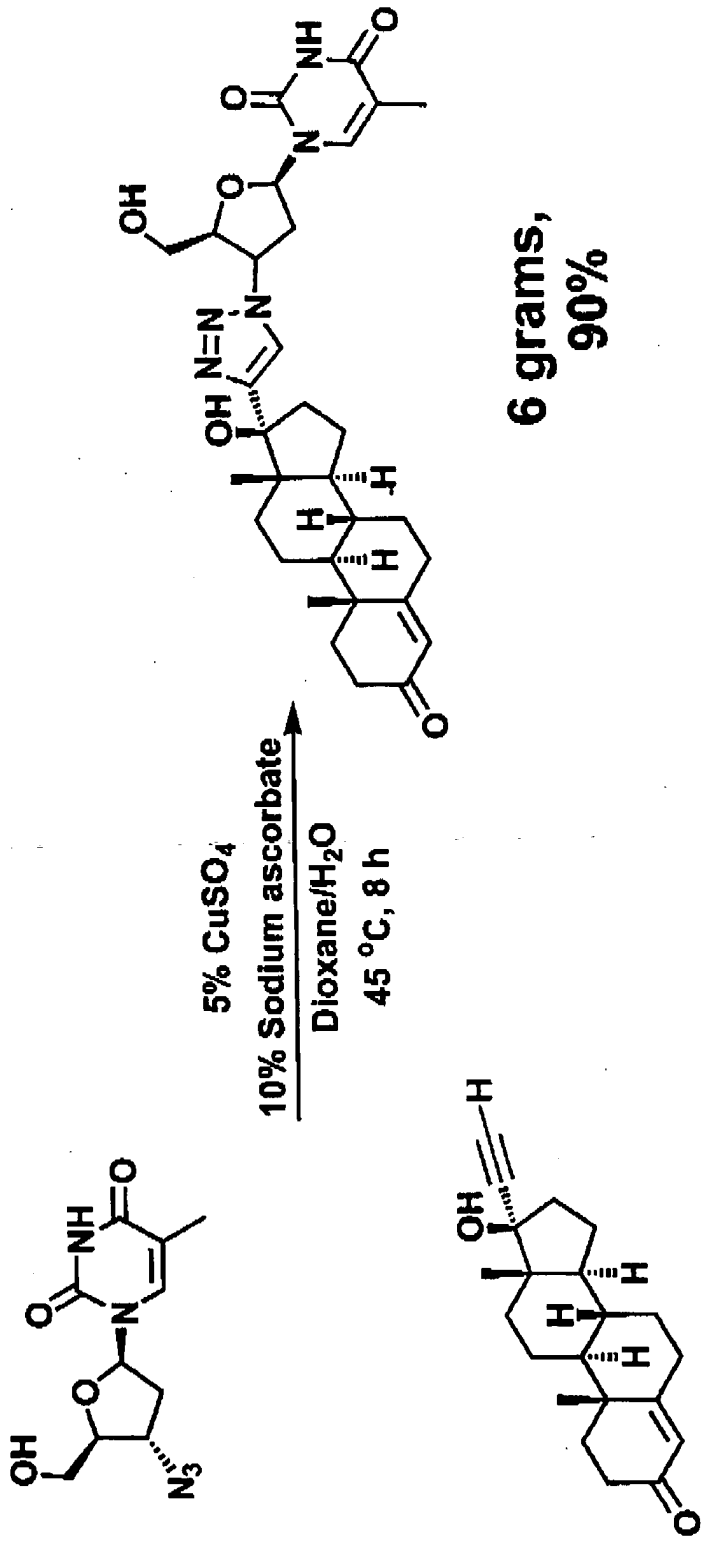
CA 100% white powder!

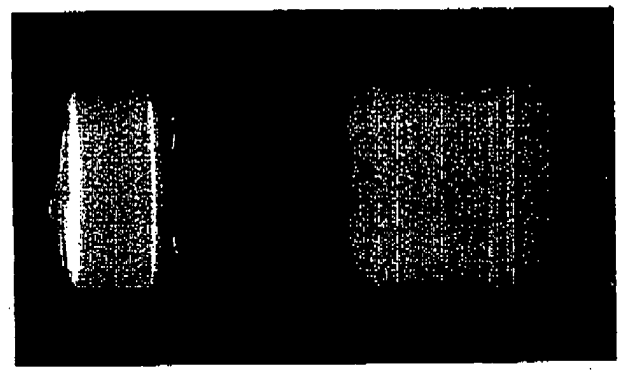
Valery

CuAAC: Amide Bond Replacement

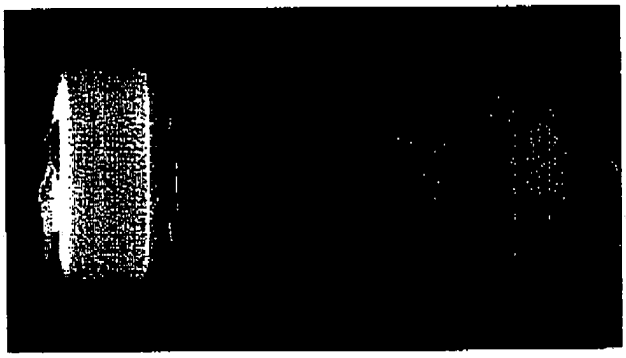


CuAAC: "Fusing" complex molecules

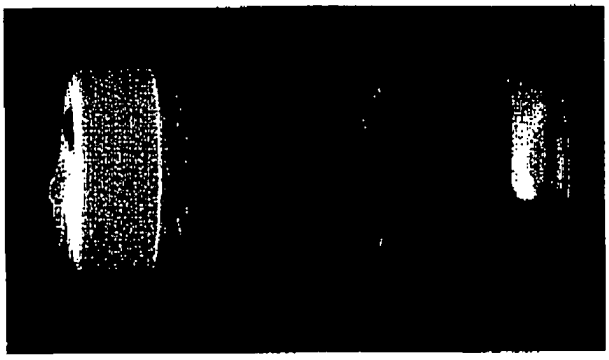




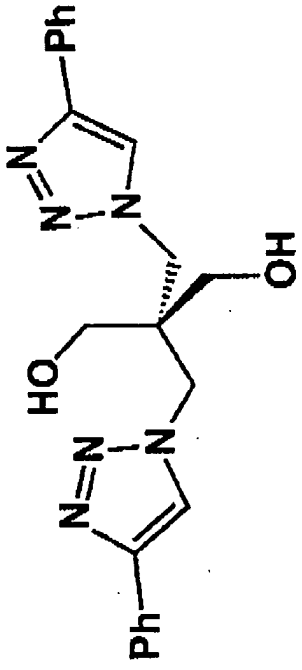
24 hrs
Completion

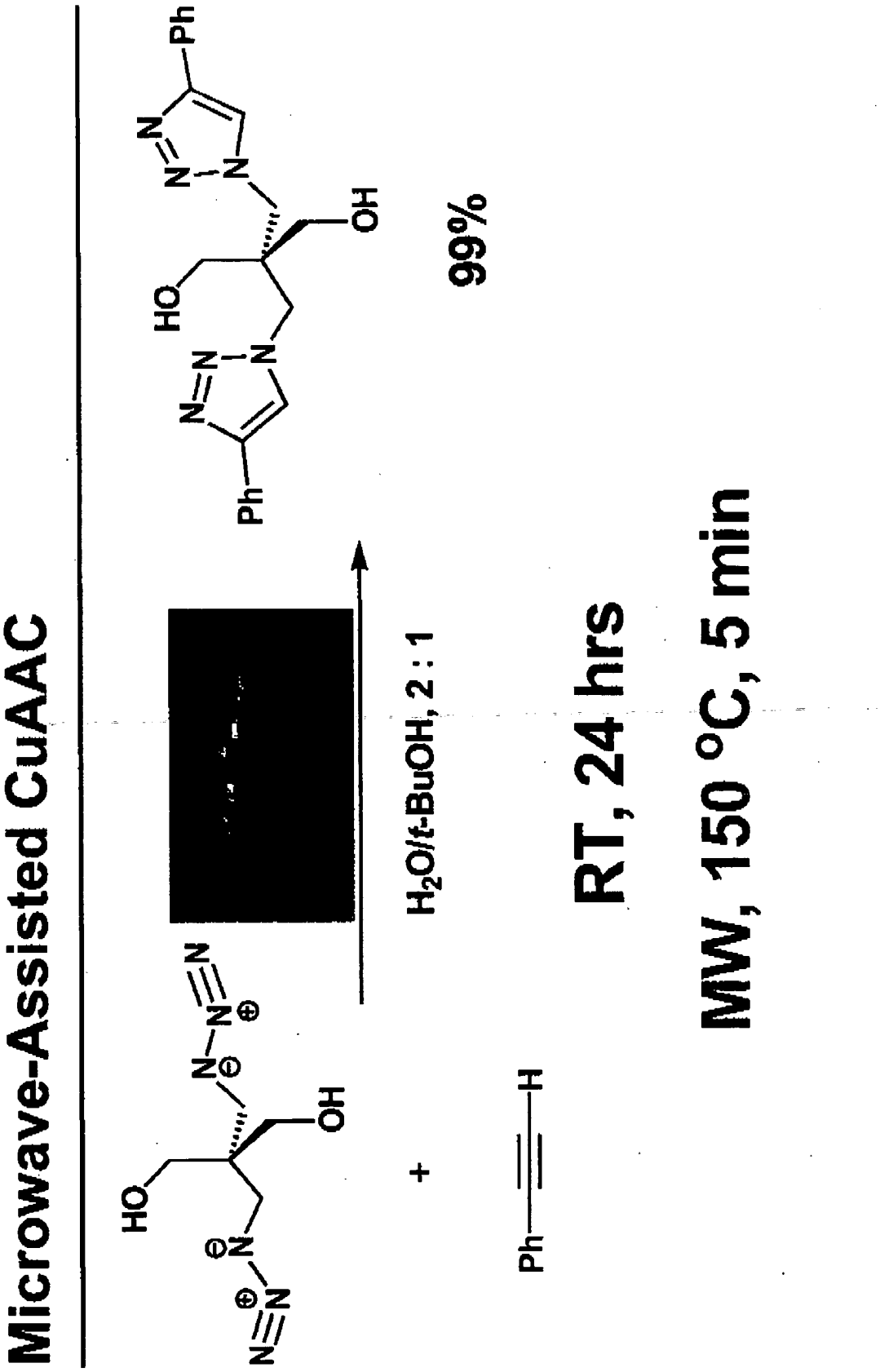


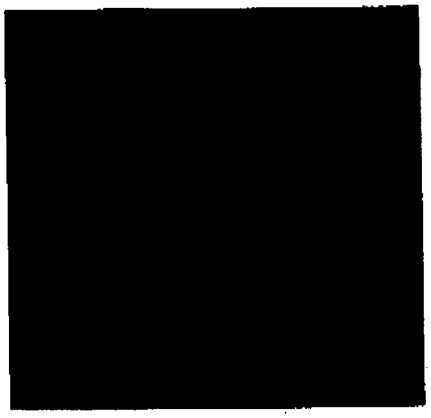
8 hrs

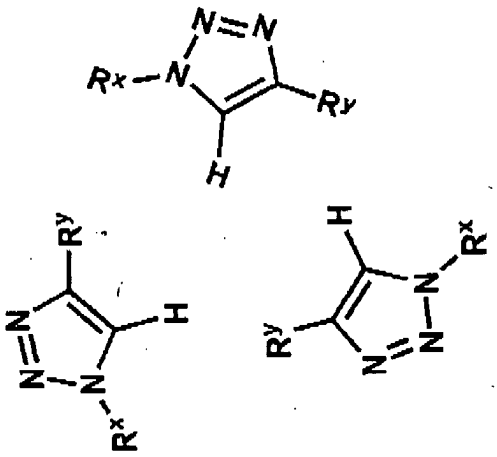


Start

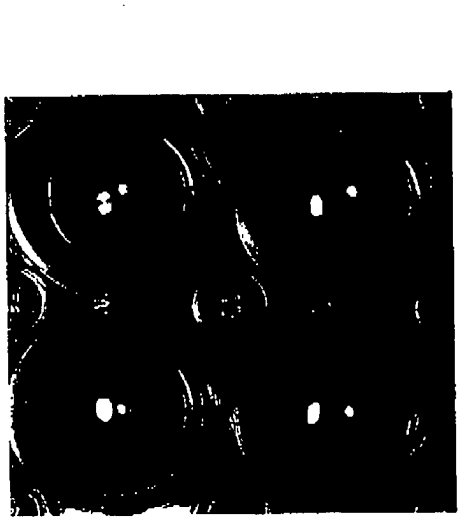


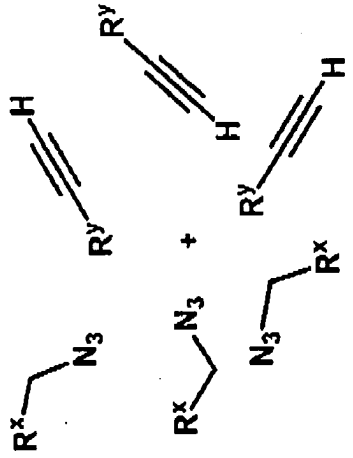

Microwave-Assisted CuAAC
Ph
N
N
N
OH
HO
N
N
N
Ph
99%
H2O/t-BuOH, 2 : 1
RT, 24 hrs
MW, 150 °C, 5 min
+
Ph
H

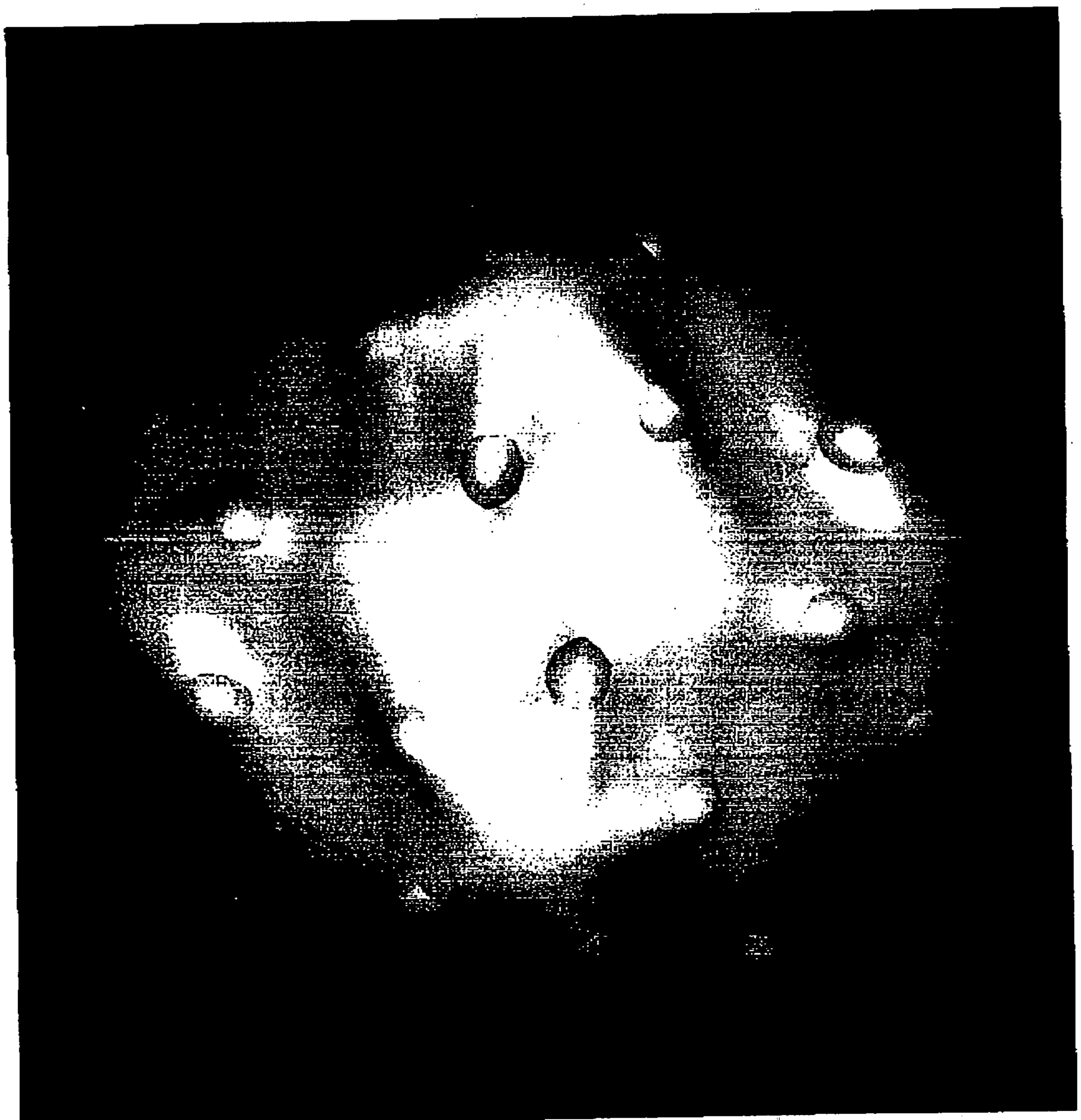




24 - 48 h
DMSO, water, ^{t}BuOH ...





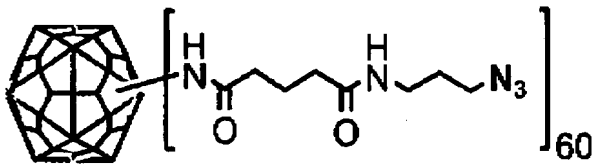


Cryo Electron Microscopy image of cysteine mutant cowpea mosaic virus labeled with monomaleimido-Nanogold.

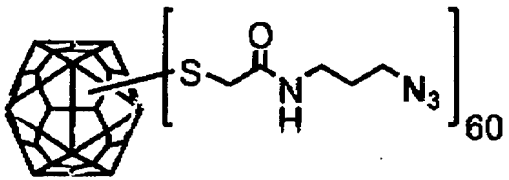
Yellow regions mark density not present in unlabeled virus, and thus the presence of the gold cluster. The position corresponds exactly with the position of the inserted cysteine residue.

M.G. Finn, J. Johnson

Bioconjugation via CuAAC



CPMV



CPMV

[virus] = 10–20 μM in total protein

160–320 nM in virus particles

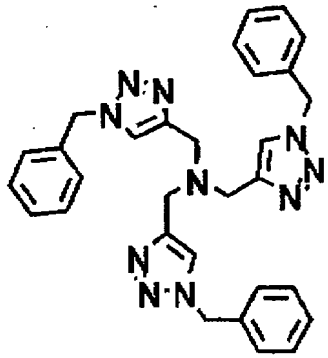
pH 7-8

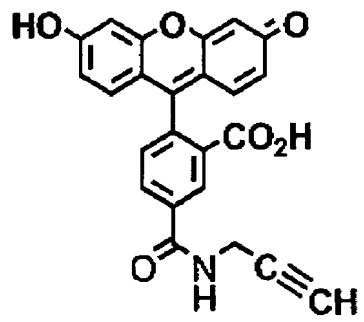
0 C

12-24 h

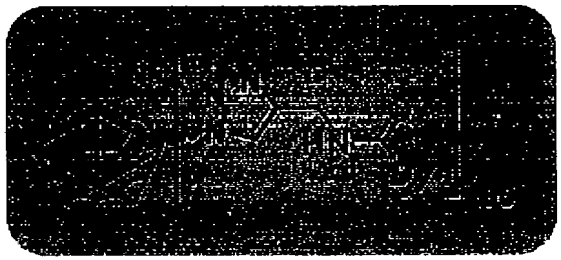
$CuSO_4$ (1 mM)

reducing agent (2 mM)





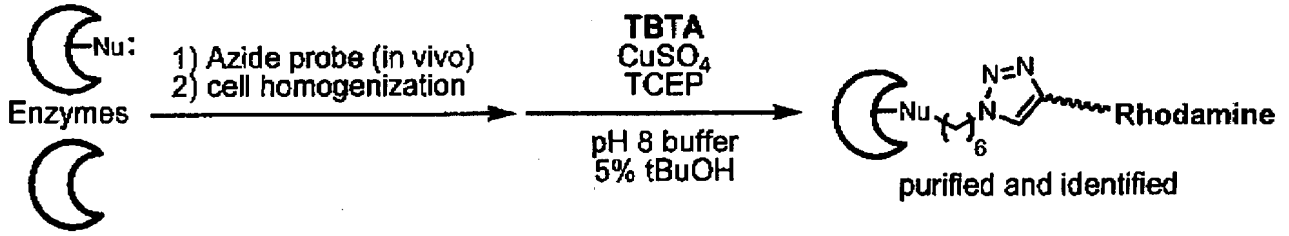
[dye] = 2 mM



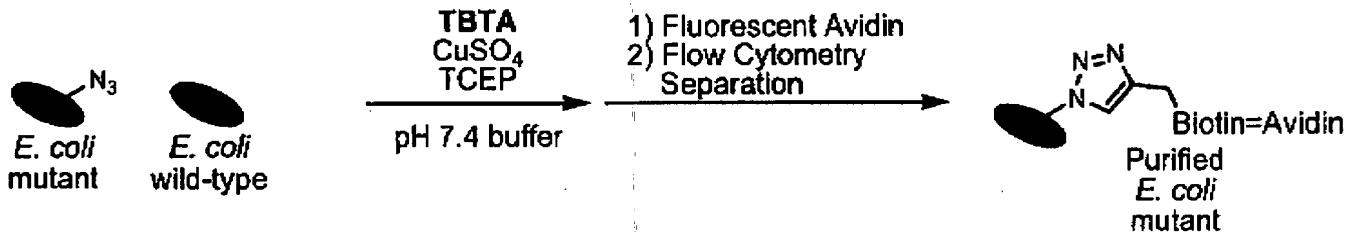
> 92%

Wang, Q., *et al.* Bioconjugation by Copper(I)-Catalyzed Azide-Alkyne [3 + 2] Cycloaddition. *J. Am. Chem. Soc.* **2003**, *125*, 3192-93

Other Examples of Bioconjugations

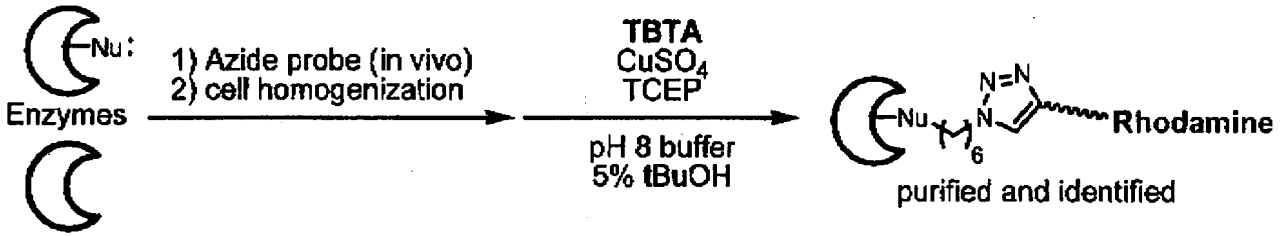


"Activity-Based Protein Profiling *in Vivo* Using a Copper(I)-Catalyzed Azide-Alkyne [3+2] Cycloaddition". Speers, A.E.; Adam, G.C.; Cravatt, B.F. *J. Am. Chem. Soc.* **2003**, *125*, 4686-4687.



"Cell Surface Labeling of *Escherichia coli* via Copper(I)-Catalyzed [3+2] Cycloaddition". Link, J. A.; Tirrell, D. A. *J. Am. Chem. Soc.* **2003**, *125*, 11164-11165.

Other Examples of Bioconjugations



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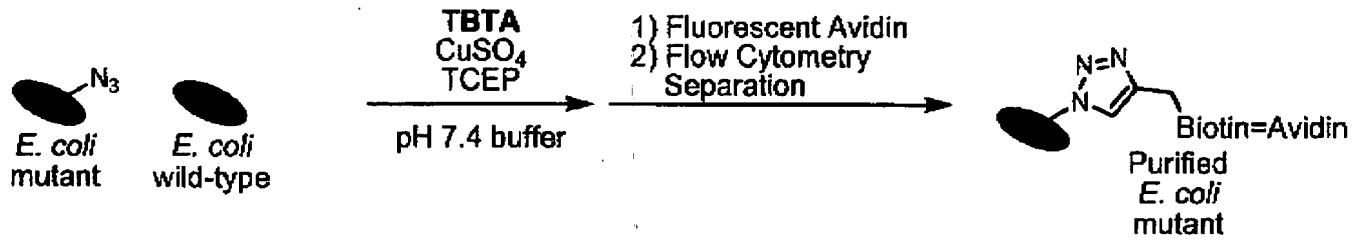

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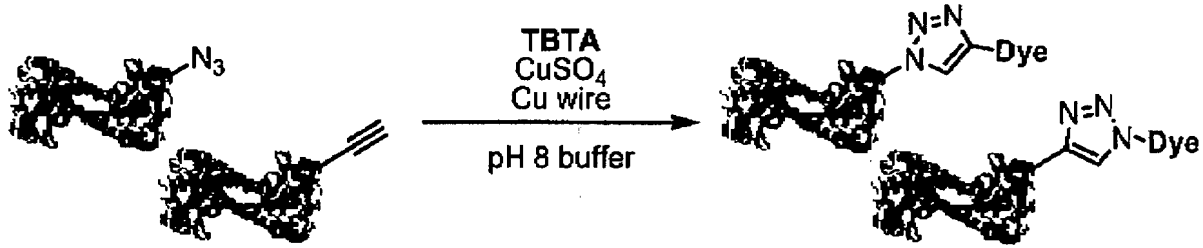
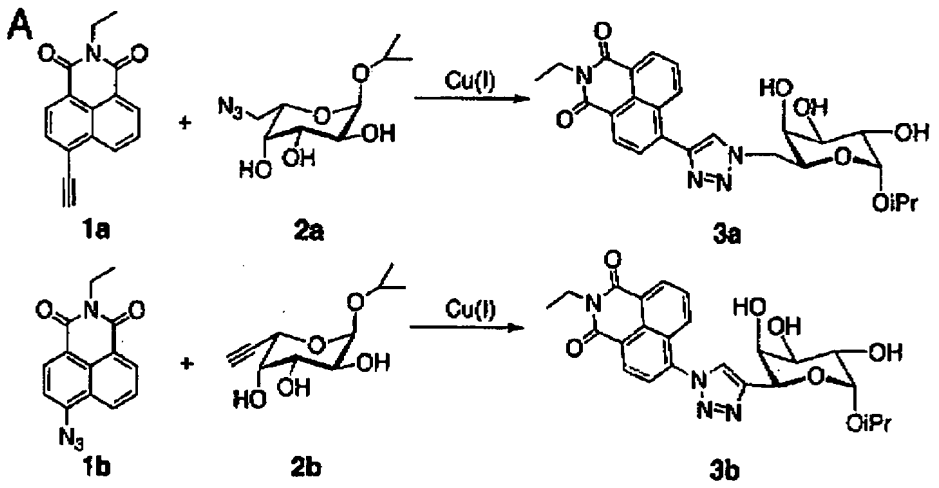
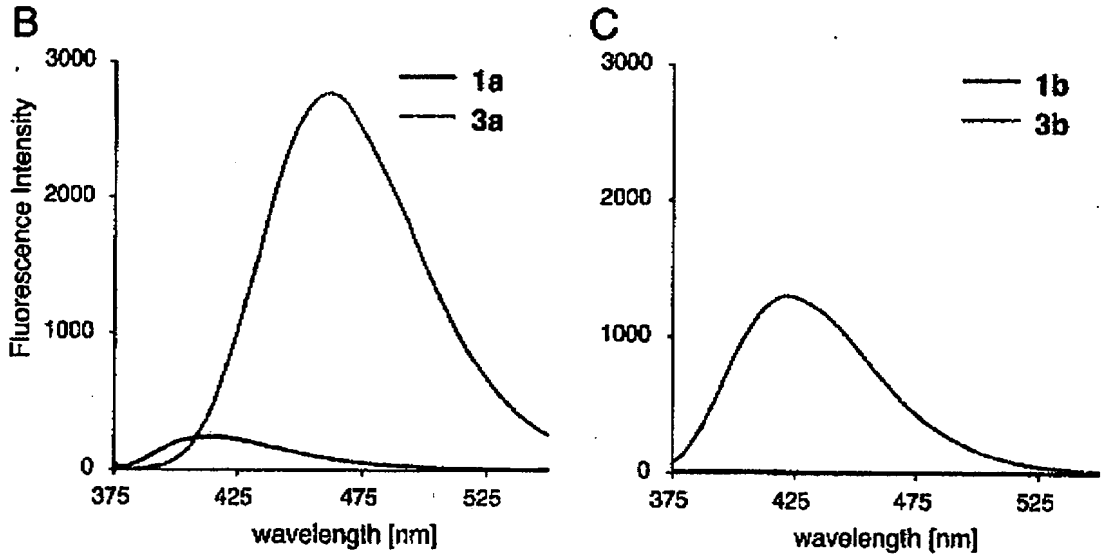


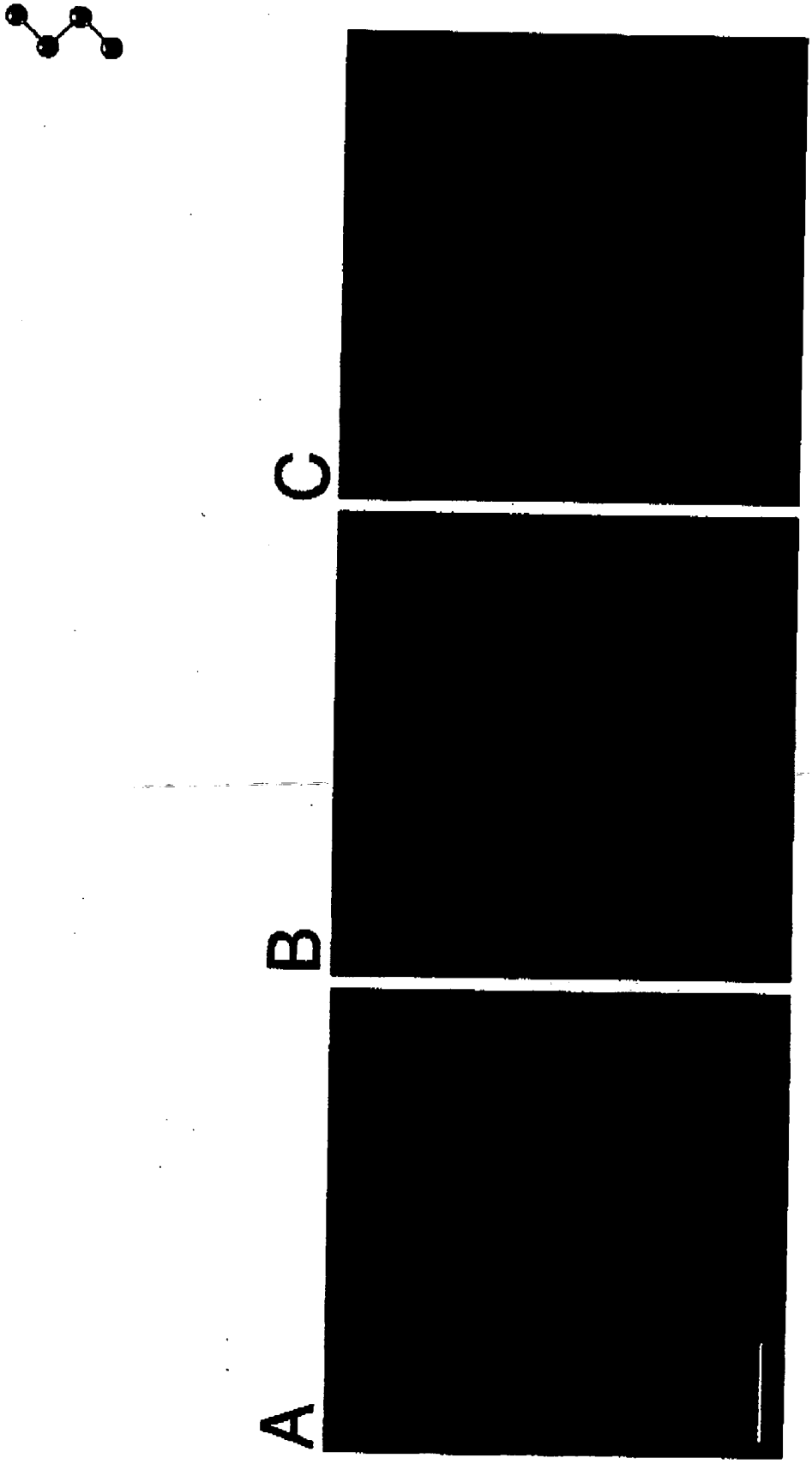
"Adding Amino Acids with Novel Reactivity to the Genetic Code of Saccharomyces Cerevisiae". Deiters, A.; Cropp, T. A; Mukherji, M.; Chin, J.W.; Anderson, J.C., Schultz, P.G.; *J. Am. Chem. Soc.* **2003**, 125(39), 11782-11783.

Glycoproteomic probes for fluorescent imaging of fucosylated glycans *in vivo*

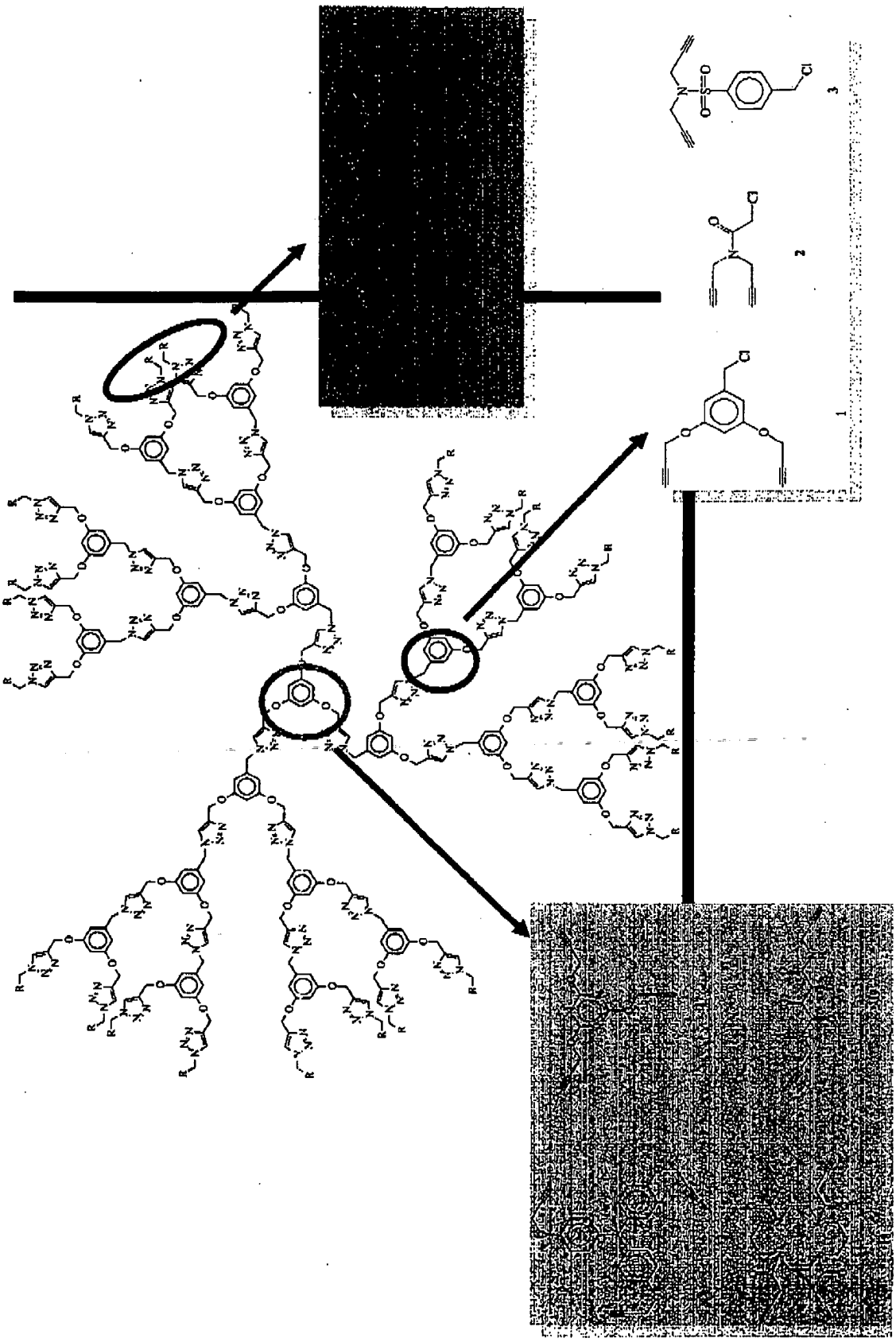
M. Sawa, T.-L. Hsu, T. Itoh , M.Sugiyama, S.R. Hanson, P.K. Vogt , and C.-H. Wong



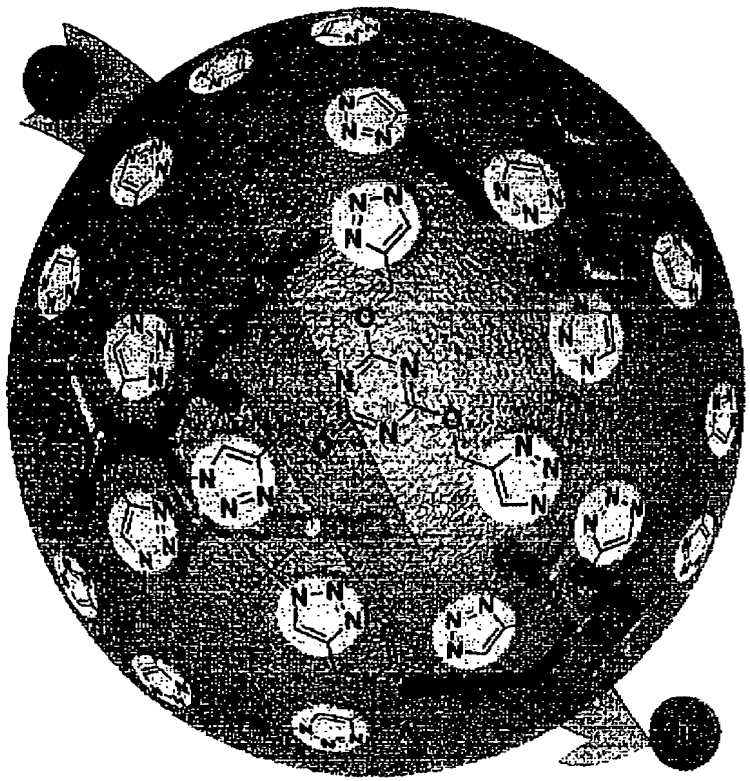



C
B
A

CuAAC: Synthesis of Dendrimers



Copper Catalysis: Synthesis of Dendrimers

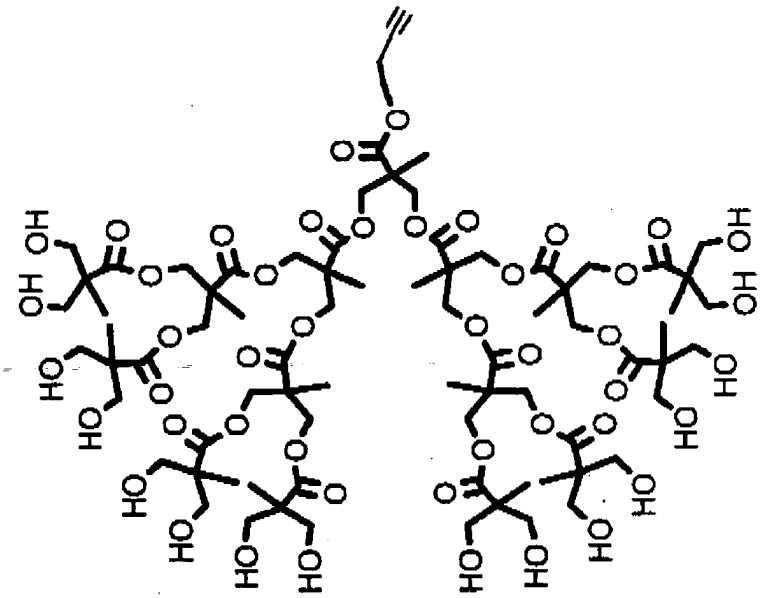


- *ca.* quantitative yield for each step of the reaction sequence
- only stoichiometric amounts of reagents
- no byproducts: greatly simplified purification

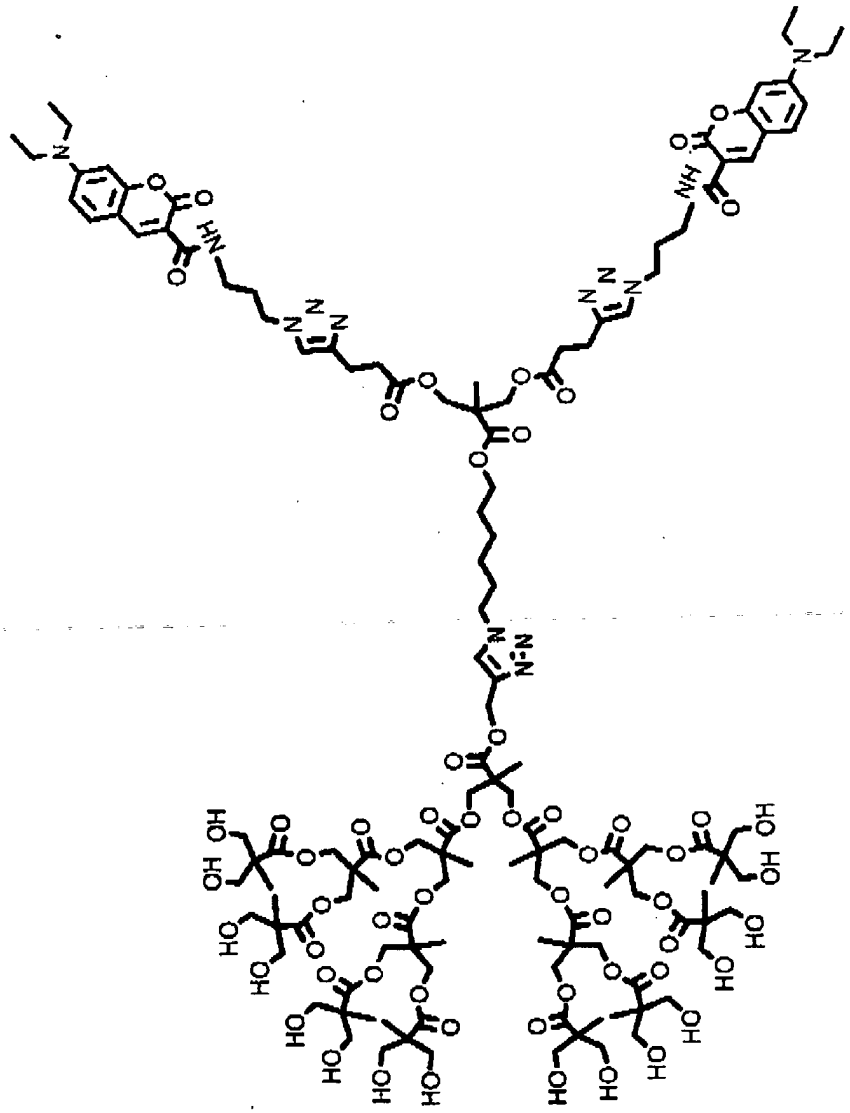
Peng Wu, Alina K. Feldman, Anne K. Nugent, Craig J. Hawker, Arnulf Scheel, Brigitte Voit, Jeffrey Pyun, Jean M.J. Fréchet, K. Barry Sharpless, and Valery V. Fokin

Angew Chem. Int. Ed. **2004**, *43*, 3863

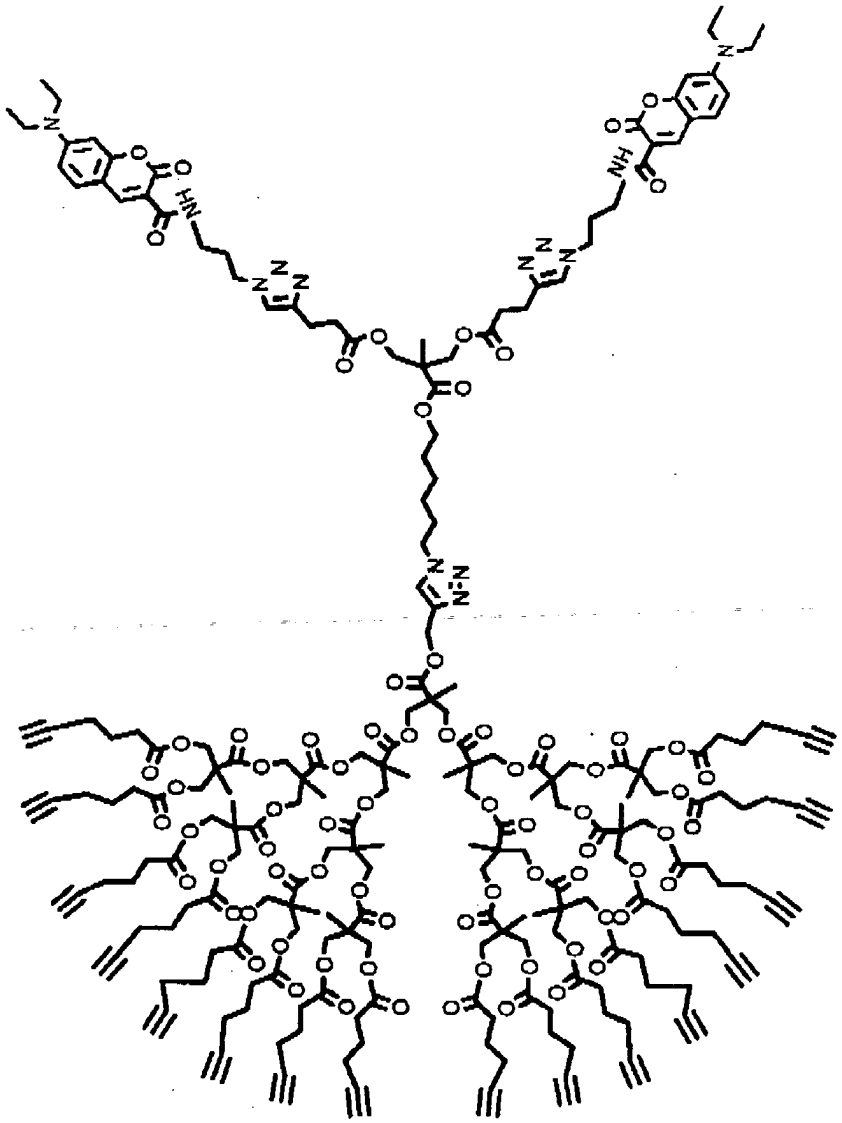
Polyfunctional Dendrimers



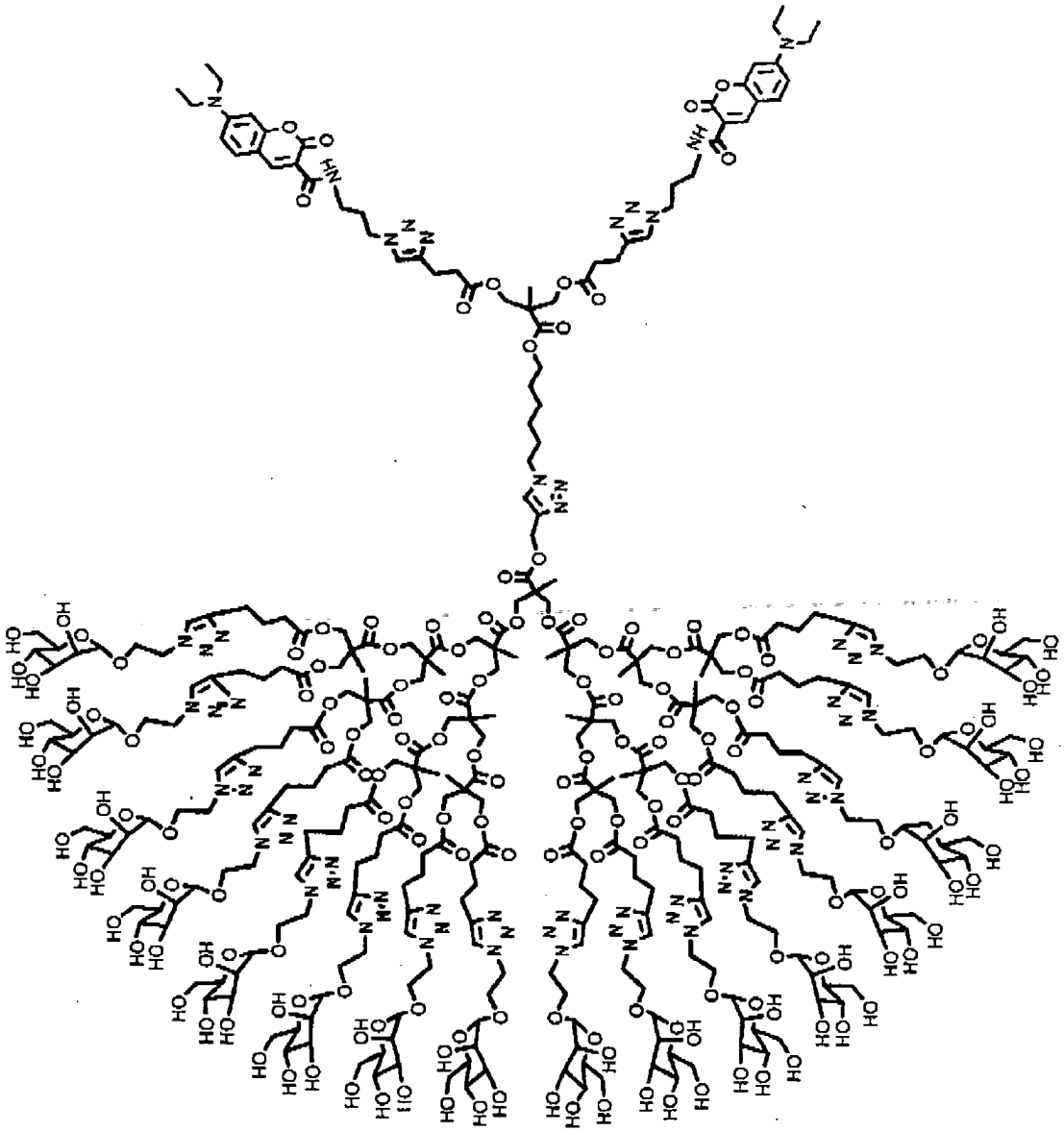
Polyfunctional Dendrimers



Polyfunctional Dendrimers

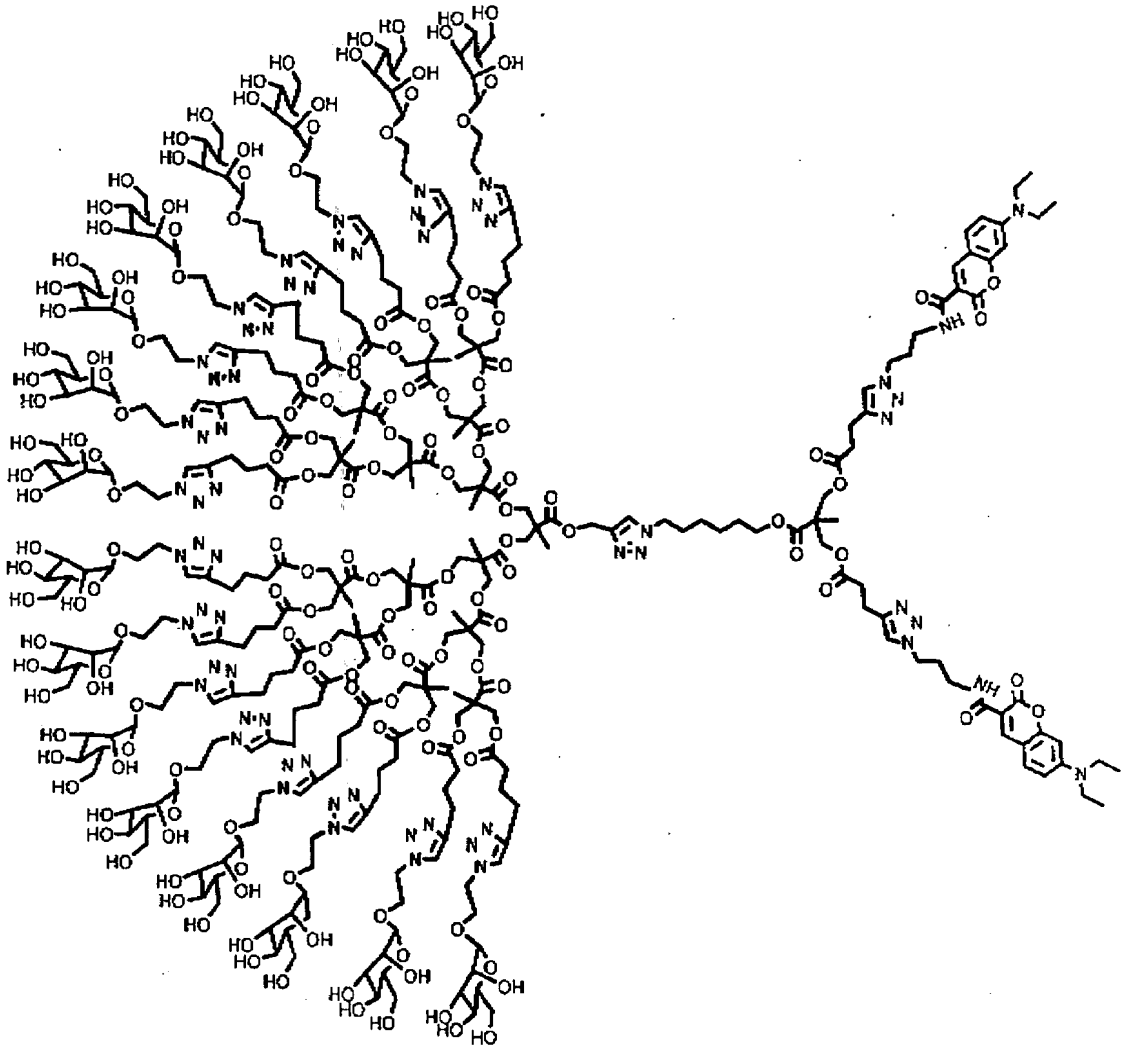


Polyfunctional Dendrimers



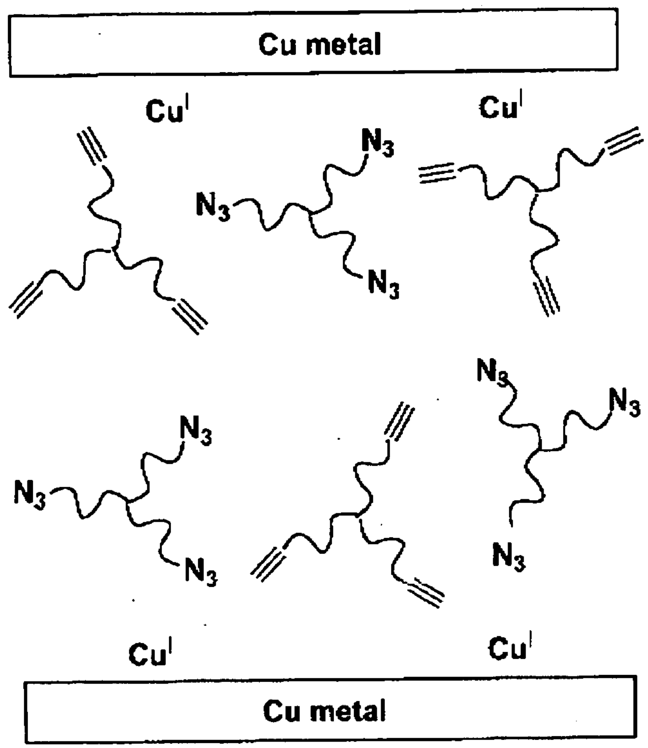
Polyfunctional Dendrimers

240-fold more potent than mannose in hemagglutination



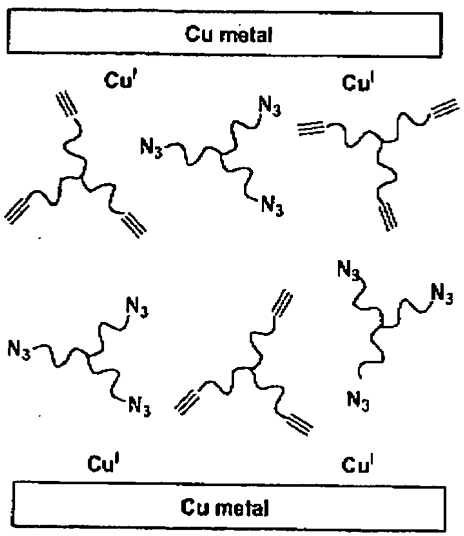
P. Wu, M. Malkoch, J. N. Hunt, R. Vestberg, E. Kaltgrad, M. G. Finn, V. V. Fokin, and C. J. Hawker, *Chem. Comm.* **2005**, 5775-5777

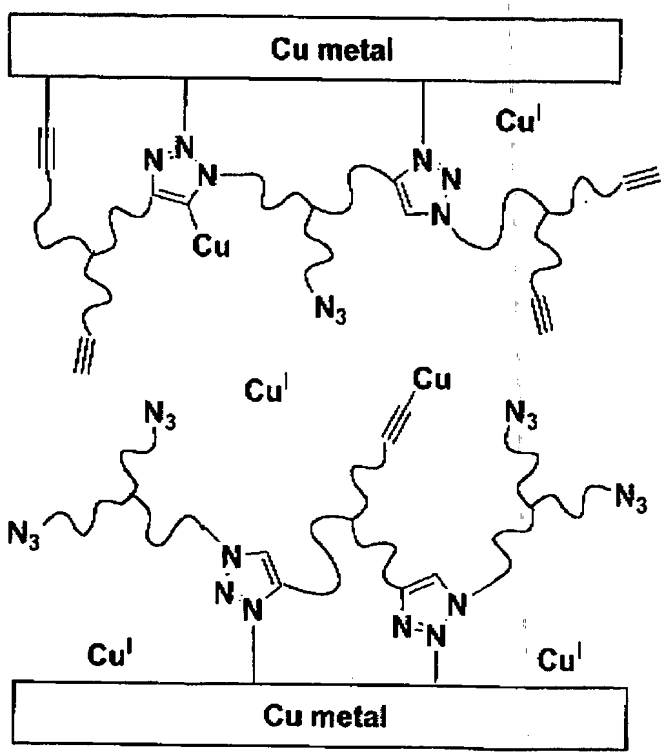
Triazole-Based Metal Adhesives



- 1,2,4-triazole containing adhesives and coatings are known.
- Cu metal has Cu^{II} and Cu^{I} ions at the surface by virtue of oxidation and comproportionation.
- Polyvalent azides and alkynes should be condensed by the Cu^{I} at the surface and thereby make an adhesive material.

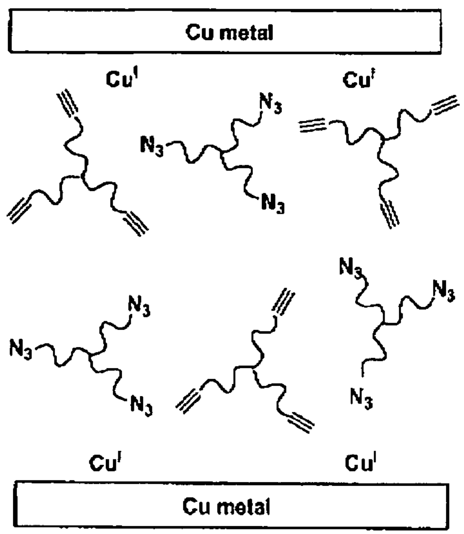
Triazole-Based Metal Adhesives

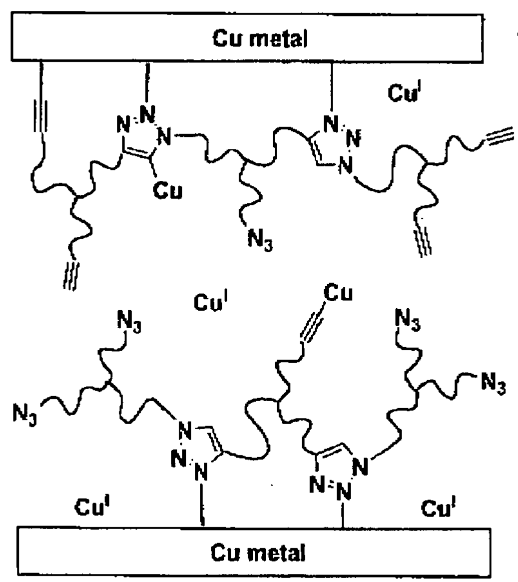


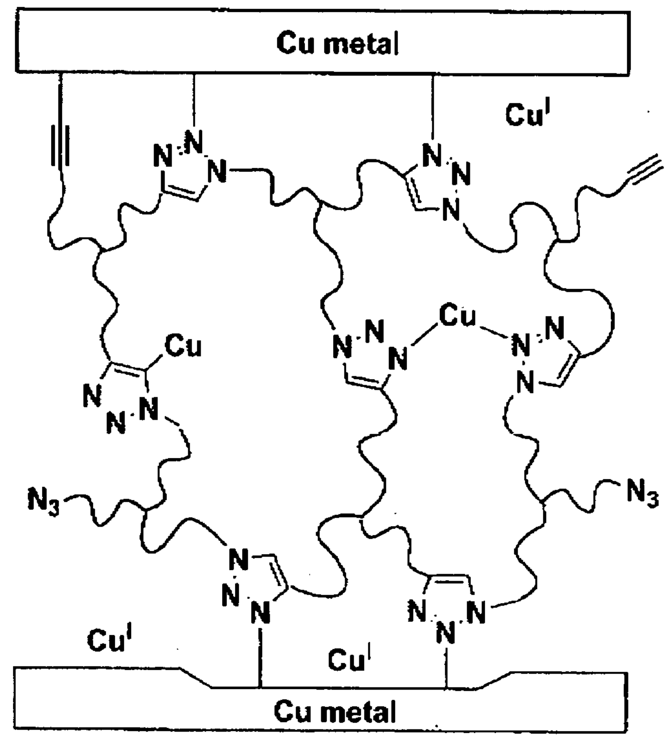


- Condensation begins at the metal surface.
- Triazole units extract or propagate Cu ions into the developing bulk polymer.

Triazole-Based Metal Adhesives





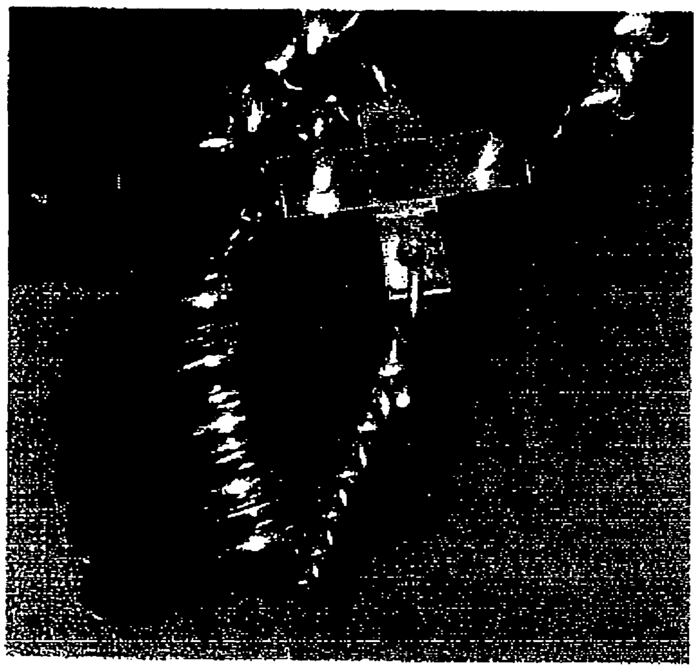


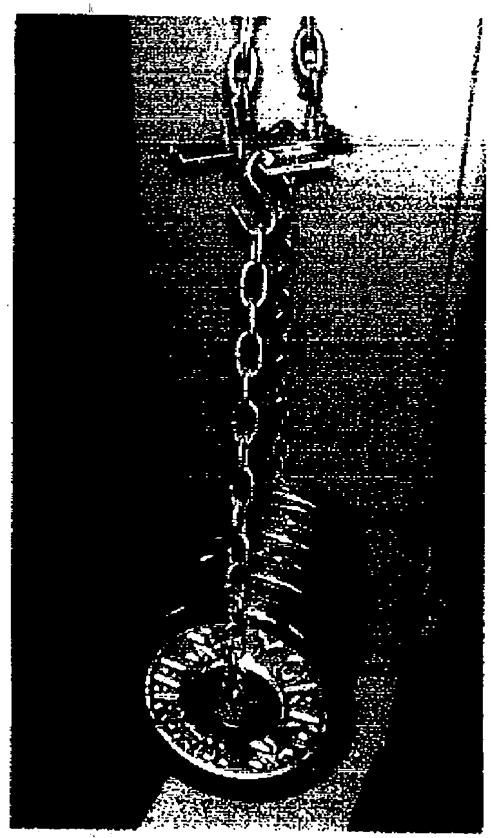
- Crosslinks form away from the metal surface as Cu ions travel with the triazole "front".
- Cu surface may be "etched" in the process.

Load Testing

- Crossed metal plates are subjected to peel-type load for constant time.
- Maximum load supported before breakage is reported.
- It is unsophisticated, but it works (error ± 1 kg on independent repetition).
- Results are reproduced well with standard instrument (H. Brown, U. Woolongong)

A

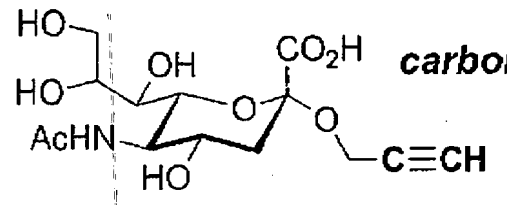




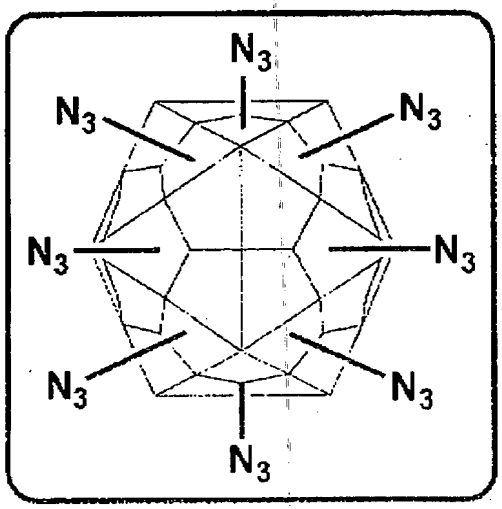
C

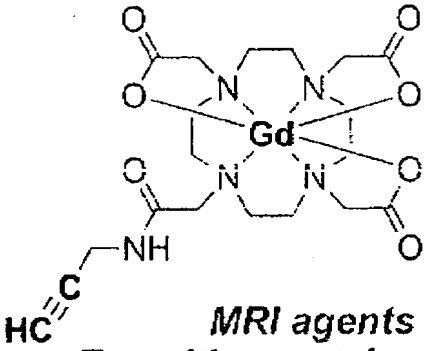
A Molecular "USB Plug" – Connecting Micro and Macro Pieces, Anywhere Any time

[redacted]



carbohydrates

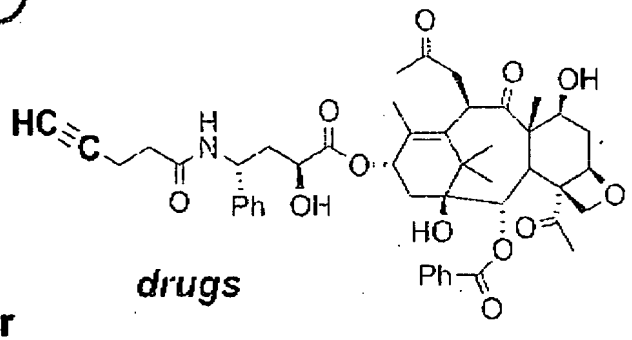




MRI agents
Transition metal catalysts

[redacted]

Also:
- **stuff to surfaces**
- **stuff to polymers**
- **stuff to agarose**
- **monomers to each other**



drugs